

FEEDING REACTIONS IN THE CILIATE, *DILEPTUS GIGAS*, WITH SPECIAL REFERENCE TO THE FUNCTION OF TRICHOCYSTS.

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I. INTRODUCTION.

Food-getting is the first necessity of the living thing, and the chief end toward which the fundamental structures of the body are directed, and this, whether in the highest mammal or lowest protozoan, becomes the chief economic problem to be solved. "Food-getting, therefore, more than any other function of the body, has been the most influential in leading to morphological development." Thus wrote one of the leading investigators in the field of protozoölogy (Calkins, '10).

¹The work here presented was largely done at the Zoölogical Laboratory of the Johns Hopkins University, Baltimore, Md., but it has been amplified by work done at the Zoölogical Laboratory of Washington University, St. Louis, Mo., and at the Marine Biological Laboratory, Woods Hole, Mass. To the directors of these laboratories I am very grateful for facilities offered.

The feeding reactions of animals have long been a favorite study for students of animal behavior. More recently the reactions of certain protozoa have been intensively studied with the hope that such problems as the choice of food would be reduced to their simplest terms in these unicellular animals which in other respects apparently stand near the bottom of the scale of development.

Ehrenberg ('38) was undoubtedly one of the first to make observations on the ability of protozoa to choose their food. He records experiments on the ingestion of carmine by various organisms in an attempt to show that protozoa do not select their food. Entz ('88) and others of his time were, however, strong in their contention that infusoria are able to select their food, ingesting certain kinds and rejecting others in a systematic way. Bütschli ('89) supports Ehrenberg's view. He concludes that protozoa do not possess the power of choice, and Verworn ('89) likewise concludes, on the basis of rather extensive experiments, that there is no selection.

Jennings ('02) states that *Vorticella* and *Stentor* probably do not have the power of selecting their food in any precise way. Mast ('09) in his work on *Didinium* shows that the apparent choice of food on the part of this organism is due to the fact that the seizing organ will adhere to the surface of some organisms and not to others. The didinia come in contact with all sorts of objects in their random swimming and "select" as food only those to which the seizing organ will adhere.

Schaeffer ('10) in his work on *Stentor coerulesus* concludes that this organism exercises a very definite selective power and discriminates very accurately between organisms and indigestible particles, and that it discriminates even between different organisms. He contends that it selects its food on a tactual basis, and apparently not on a chemical one. The same worker ('16) reports some experiments on *Amoeba* and maintains (p. 562) that although *Amoeba* eats insoluble substances, there is a slow process of learning in favor of selection. He ascribes to the endoplasm of *Amoeba* a more specific power of discrimination than to the ectoplasm, and also maintains that movement of an object is a very important factor in determining whether or not it shall be eaten.

Calkins ('10) says, "while most of the protozoa wait until the

prey comes to them, and take what they can get, others are predatory and go in search of food. These are the most interesting of all protozoa, for they are occasionally too fastidious apparently to take the ordinary run of microscopic wilds, but seem to select their food with all the care of a gourmand." As an example of this type he describes the reactions of *Actinobolus radians*.

Miss Moody ('12) in her study of *Actinobolus* and *Spathidium* asserts that they "subsist exclusively on a special type of ciliate. *Actinobolus* awaits the coming of *Halteria grandinella* before making use of its weapons of offense," while *Spathidium* swims about "with seeming indifference to all food material except the little ciliate, *Colpidium colpoda*." She concludes that "the protoplasm of these organisms has become modified chemically and physiologically to such an extent that a reaction to one kind of protoplasm only is possible; in other words, forms like *Actinobolus* and *Spathidium* have become "educated through 'error' to the selection of one species of food each, namely, *Halteria grandinella* and *Colpidium colpoda*."

Metalnikow ('12) contends that if paramecia are fed for some time on a non-digestible substance, they take in gradually less and less, until finally they refuse it entirely under all conditions, but that they nevertheless take in other substances just as before. He shows that in the case of feeding on carnine this power of selection is lost at the time of division. He also shows that there is a decided power of discrimination between substances already within the body; for some substances are quickly excreted, while others remain within the body for a considerable length of time.

In his studies of one of the Suctoria, *Podophrya collini*, Root ('14) maintains that there are several definite factors which determine the selection of food in this organism. He shows that the character of the outer surface of certain organisms as to physical and chemical constitution, mucus secretion, etc., prevents the attachment of the seizing apparatus. He shows, moreover, that the size, the activity, and the characteristic behavior of certain organisms in relation to the sessile habits of *Podophrya collini* are also determining factors.

From this brief review of some of the more general literature in this field it is evident that selection of food has thus far been

positively demonstrated in only a very few forms, while in general it would appear that most workers have supported the opposing view.

There are only a few incidental references in the literature to the feeding habits of *Dileptus gigas*. Bütschli ('89) says its food is "sehr grob," and is quoted by Calkins as saying that it feeds on ciliates alone. According to Wrzesniowski ('70), "*Dileptus gigas* is a voracious animal which feeds only on living food, preying especially on *Stylonychia*." Pritchard ('61) says it feeds largely on green monads, because of which it is often of a green color. Hausman ('17) says "*Dileptus* is surely the king of beasts among the ciliated protozoa. It is entirely carnivorous and its appetite is apparently insatiable. The prey is stung by well-developed trichocysts, and if too large to be swept into the buccal cavity by the cilia, it is forced in by the writhings of the neck" (proboscis).

It is clearly evident that there are a number of different views concerning the feeding habits of this infusorian, all of which are apparently based on purely incidental observations. Does *Dileptus* feed on ciliates alone, or even on living food only, which would involve the power of choice of food? Does it paralyze its prey by means of trichocysts? What is the nature of these structures? These are the problems which are considered in the observations and experiments which comprise the material presented in this paper.

The work was begun at the suggestion of Professor S. O. Mast, to whom I am deeply indebted for many helpful suggestions concerning the experiments made and for much valuable criticism during the preparation of this paper.

2. MATERIAL AND METHODS.

Dileptus gigas is one of the holotrichous ciliates belonging to the family Tracheliidae. It is one of the largest of the more common protozoa, often measuring over 600 micra in length. It possesses an elongated body, sharply pointed at the posterior end, and at the anterior end drawn out into a long proboscis which is frequently as long as the body itself. The mouth opening is located at the basal end of this proboscis, and has a circular aperture with a short funnel-shaped gullet leading from it (Fig. 1). Both these struc-

tures are capable of enormous expansion at the time of feeding. Normally, however, they are closed except for a pit-like cavity which is always present. A cytopyge is sometimes discernible near

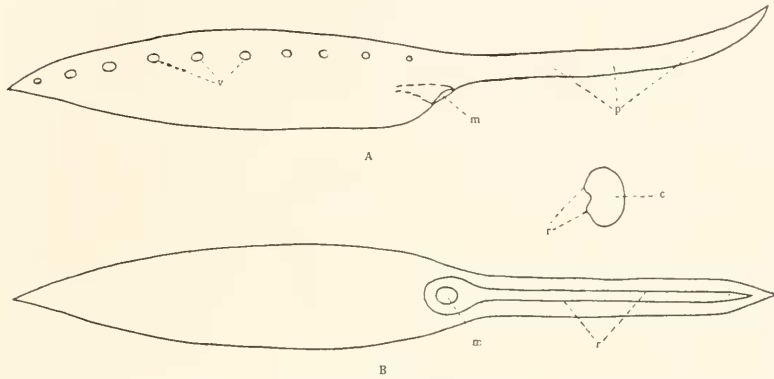


FIG. 1. Diagrammatic sketches of *Dileptus gigas*: *A*, side view; *B*, oral view; *v*, contractile vacuoles; *p*, proboscis; *m*, mouth; *c*, cross section of proboscis; *r*, bands of large cilia.

the posterior end, at which place faecal material often collects in a large vacuole from which it is sporadically discharged. There are numerous contractile vacuoles of which the larger ones are arranged in a series near the aboral surface. Thin contractile fibrillæ extend around the body in the form of a flat spiral, effecting about one complete turn for the entire length of the body. The entire body is covered with short cilia which run in rows parallel to the fibrillæ. On the ventral surface of the proboscis, which is somewhat flattened, the cilia are considerably thicker and longer than elsewhere, especially along the edges, where they are in the form of two bands. These extend backward in such a way as to meet and form an arch just behind the mouth opening. Since all these cilia beat backward under normal conditions, a decided current is produced in the groove between these two bands of large cilia. This current starts at the tip of the proboscis and runs back to the mouth, where it ends in a sort of vortex, due to the action of the band of cilia which partially surrounds this pit-like orifice as above described. Structures which have been quite generally described as trichocysts are found in the oral surface of the proboscis. They can be seen only indistinctly in living mate-

rial. There are at least several hundred of these structures, all of which are arranged in a band extending from the mouth to the tip of the proboscis, approximately in the mid-line of its oral surface.

There has been much discussion concerning the nuclear condition of this organism, but recent investigators appear to be agreed that *Dileptus* possesses a distributed nucleus. An account of this condition will be omitted here, as it is planned to present a discussion of the nuclear phenomena during the life history of this organism in a separate paper.

Dileptus is never at rest. It is always swimming, as a rule quite slowly unless disturbed. In cultures it is observed to spend most of its time swimming slowly just above the debris at the bottom of the dishes. Apparently a like condition obtains in nature, for if *Dileptus* is found in a pool, samples taken from near the bottom contain many more specimens than those taken at higher levels. It is observed to progress with its posterior end close to or in contact with the bottom, while its anterior end is always at a noticeable elevation. It proceeds with its proboscis ahead, continually waving it now this way, now that, as if in search of food. It also rotates slowly on its longitudinal axis. This rotation, in connection with the searching movements of the proboscis, enables it to explore a very large area.

In January, 1919, a few specimens of *Dileptus gigas* appeared in an old "paramecium-culture" in which active fermentation had long since ceased. It can not live long in any culture in which active fermentation is taking place, nor can it thrive in cultures rich in organic food supply. Consequently, very dilute infusions were made, and these were usually inoculated with *Euglena gracilis* before introducing *Dileptus*. In this manner the original culture was kept for more than fifteen months.

In nature I have found them in the large pond on the Homewood Campus of the Johns Hopkins University, Baltimore, and also in the "school-house" pond near the Biological Laboratory at Cold Spring Harbor, L. I. They were found also in the Cedar Swamp pond at Woods Hole and in the outlet channel of Creve Cœur Lake near St. Louis, Mo. In all these ponds at the time *Dileptus* was found there was but little organic decay taking place and the water was "relatively pure."

In preparation for the experiments described in the following pages, a number of the organisms, usually about fifteen, were transferred with a capillary pipette from the stock-culture to small dishes containing about 5 c.c. of spring water. They were left in this water without food for from 2 to 3 days. During this starvation process about ten per cent. usually encysted and the rest diminished considerably in size, became very hungry, and were consequently in excellent condition for observation on the selection of food and the process of feeding. When kept longer, without food, they continued to diminish in size until in the course of a week they were reduced to less than one tenth of their former length and probably to less than one one-hundredth of their former volume, after this they disintegrated unless this was prevented by opportune feeding.

All the experiments on the selection of food were conducted as follows: A designated number of starved dilepti, usually fifteen, were placed in a small watch-glass with about 3 c.c. of spring water. A drop of a concentrated suspension of the substance to be tested was then added and the whole thoroughly mixed. The feeding reactions of some few of the specimens were carefully observed for longer or shorter periods of time. At the end of twenty minutes all the dilepti were taken out of this medium by means of a small pipette and introduced into a like volume of spring water. They were then observed, either individually or two at a time, on a slide under a magnification of about 300 diameters, and the number of vacuoles in each was carefully counted. In this way various living organisms and numerous inanimate particles were tested.

The protoplasm of *Dileptus* is quite opaque under ordinary conditions of culture. Occasionally a culture was obtained in which the organisms were relatively transparent and it was only with such material that the feeding experiments were made, for even under optimum conditions there was some difficulty in seeing precisely what passed into the body unless it was specifically colored as, *e.g.*, carmine or india ink. Another factor which favored observation on the amount of food ingested was the characteristic large size of the first vacuoles formed, whenever an organism was introduced into a new medium, especially after a period of starvation. Such large vacuoles are very specific.

3. EXPERIMENTS ON SELECTION OF FOOD.

A. *Inanimate Substances.*

In experiments on inanimate substances it was desired to use only insoluble and non-toxic materials. The following substances were tested: carmine, chalk, sand, powdered glass, and india ink. Three of these, namely, carmine, glass, and india ink, appeared to be more favorable than the others, because of the fact that vacuoles filled with these substances were readily distinguishable. The results obtained in ten experiments with each of these three substances are given in Tables I., II., and III.

Table I. contains the results obtained with carmine. This table

TABLE I.

EXPERIMENTS ON CARMINE.

Table showing results obtained in feeding *Dileptus* on carmine. In the five columns under the headings 0-4, are indicated the number of individuals which formed 0, 1, 2, 3, 4, or more vacuoles containing carmine, respectively, during the twenty minutes of the experiment. Carmine was ingested by 97.5 per cent. of the individuals but in only 17 per cent. was there more than one vacuole containing carmine formed.

Experiment Number.	Total Number of Dilepti.	Number of Carmine Vacuoles.				
		0	1	2	3	4 or More.
1.....	15	0	14	1	0	0
2.....	15	0	12	2	1	0
3.....	15	0	8	7	0	0
4.....	14	0	11	3	0	0
5.....	15	1	13	1	0	0
6.....	15	1	12	2	0	0
7.....	13	0	12	1	0	0
8.....	15	1	11	2	1	0
9.....	16	0	13	3	0	0
10.....	16	0	14	1	1	0
Average.....	14.9	.3	12	2.3	.3	0
Per cent.....		2	50.5	15	2	-

shows that only three of the one hundred and forty-nine individuals tested did not ingest carmine, that more than eighty per cent. of the total number tested formed one vacuole, while fifteen per cent. formed two vacuoles each. Thus it is evident that carmine is eaten, but only in small quantities, for in only seventeen

per cent. of the individuals tested was there more than one vacuole containing carmine formed.

Table II. contains the results of ten experiments with powdered

TABLE II.

EXPERIMENTS ON BLUE GLASS

Table showing the results obtained in feeding *Dileptus* on powdered blue glass. In the five columns under the headings 0-4, are indicated the number of individuals which formed 0, 1, 2, 3, 4, or more vacuoles containing blue glass, respectively, during the twenty minutes of the experiment. Glass was ingested by 89.3 per cent, but in only 12 per cent. was there more than a single vacuole containing glass formed.

Experiment Number.	Total Number of Dilepti.	Number of Vacuoles Containing Glass.				
		0	1	2	3	4 or More.
1.....	15	4	8	2	1	0
2.....	14	3	10	1	0	0
3.....	15	0	10	3	1	1
4.....	15	0	15	0	0	0
5.....	15	1	14	0	0	0
6.....	15	3	9	2	0	1
7.....	16	0	16	0	0	0
8.....	15	0	14	1	0	0
9.....	15	4	11	0	0	0
10.....	15	1	9	3	1	1
Average.....	15	1.6	11.6	1.2	.3	.3
Per cent.....		10.7	77.3	8	2	2

blue glass. It shows that eighty-nine per cent. were observed with vacuoles containing glass; that although so large a percentage had ingested glass to some degree, yet only twelve per cent. formed more than one vacuole; and that only two per cent. were observed with more than three vacuoles containing glass. It is therefore evident that while glass is ingested, it is taken only in very small amounts.

Table III. contains the results obtained with india ink. It shows that almost ninety-five per cent. contained at least one vacuole with ink particles in it, and that only twenty-seven per cent. had formed a second vacuole during the entire twenty minutes. We can conclude, therefore, that ink is eaten in small amounts by the great majority of specimens of *Dileptus*, but that only relatively few form more than two vacuoles containing this substance.

TABLE III.

EXPERIMENTS ON INDIA INK.

Table showing results obtained in feeding *Dileptus* on India ink. In the columns under the headings of 0-4, are indicated the number of individuals which formed 0, 1, 2, 3, 4, or more vacuoles each, containing India ink, during the twenty minutes of the experiment. Ink was ingested by more than 94 per cent., but less than 28 per cent. formed more than a single vacuole containing ink.

Experiment Number.	Total Number of <i>Dilepti</i> .	Number of Vacuoles Filled with Ink.				
		0	1	2	3	4 or More.
1.....	15	1	11	2	0	1
2.....	15	0	12	2	1	0
3.....	15	0	11	1	2	1
4.....	16	1	13	2	0	0
5.....	17	2	11	3	1	0
6.....	15	3	12	0	0	0
7.....	15	0	8	4	1	2
8.....	12	1	7	3	0	1
9.....	15	0	3	5	4	3
10.....	15	0	13	2	0	0
Average.....	15	.8	10.1	2.4	.9	.8
Per cent.....		5.3	67.3	16	6	5.3

In the experiments on all the other inanimate substances mentioned earlier, results were obtained which are, in the main, in harmony with those presented in Tables I., II., and III. All these substances, with the possible exception of sand, were ingested by a great majority of the individuals used in the tests. Experimentation with sand was very difficult owing to the fact that it settles very quickly, and that it is also difficult to see. My notes on the few experiments made record only thirty-two per cent. as having fed on this substance. The results obtained in experiments with chalk are almost identical with those obtained with glass. In the experiments on starch there does not appear to have been as sharp a decline between the number forming only one vacuole and those forming three or four vacuoles each within the twenty minutes. In other words, the power of discrimination does not seem to be as well developed in regard to this substance as it is in regard to the others. These experiments on inanimate substances thus show quite clearly that *Dileptus* when hungry will ingest insoluble substances, but that usually only one vacuole is formed.

B. *Animate Substances.*

In comparison with the results obtained in the above-described experiments on inanimate substances, those obtained in experiments on living material stand out in sharp contrast. Experiments were

TABLE IV.

EXPERIMENTS ON *Euglena*.

Table showing results of ten experiments obtained in feeding *Dileptus* on *Euglena*. In the five columns under the headings 0-4, are indicated the number of individuals which formed, in twenty minutes, 0, 1, 2, 3, 4, or more vacuoles containing *Euglena*. *Euglena* was ingested by more than 94 per cent. and a second vacuole containing this flagellate was formed by 89.3 per cent. of the individuals tested.

Experiment Number.	Total Number of Dilepti.	Number of Vacuoles Containing <i>Euglena</i> .				
		0	1	2	3	4 or More.
1.....	15	1	0	1	4	9
2.....	15	0	0	1	3	11
3.....	14	0	0	1	5	8
4.....	12	1	2	0	4	5
5.....	16	0	1	2	2	11
6.....	17	0	1	3	2	11
7.....	15	0	0	1	5	9
8.....	15	0	0	0	3	12
9.....	16	6	3	2	3	2
10.....	15	0	1	3	2	9
Average.....	15	.8	.8	1.4	3.3	8.7
Per cent.....		5.3	5.3	19.3	22	58

made with most of the forms listed in Table VII. The results obtained with all of these forms are essentially the same. Those obtained with *Euglena*, *Colpidium*, and *Chilomonas* are presented in Tables IV., V., and VI. By referring to these tables it will be seen that *Euglena* was ingested by 95 per cent. of the dilepti tested, 89 per cent. forming three or more vacuoles, and 50 per cent. four or more; that *Chilomonas* was ingested by 98 per cent. of the dilepti tested, 70 per cent. forming three or more vacuoles; and that *Colpidium* was ingested by 99 per cent. of the dilepti tested, 84 per cent. forming more than one vacuole. It is consequently evident that these organisms are ingested in large numbers by the majority of the dilepti tested.

TABLE V.

EXPERIMENTS ON *Colpidium*.

Table showing results obtained in feeding *Dileptus* on *Colpidium*. In the five columns under the headings 0-4, are indicated the number of individuals which formed, in twenty minutes 0, 1, 2, 3, 4, or more vacuoles containing *Colpidium*. *Colpidium* was ingested by 99 per cent. and 84 per cent. formed more than one vacuole.

Experiment Number.	Total Number of Dilepti.	Number of Vacuoles Containing <i>Colpidium</i> .				
		0	1	2	3	4 or More.
1.....	14	0	3	5	6	0
2.....	15	0	2	4	3	6
3.....	12	0	3	2	5	2
4.....	11	0	0	4	6	1
5.....	15	1	5	3	4	2
6.....	15	0	1	7	2	5
7.....	12	0	3	4	4	1
8.....	12	0	2	3	6	1
9.....	15	0	0	4	8	3
10.....	14	0	1	3	7	3
Average.....	13.5	.1	2	3.9	5.1	2.4
Per cent.....		.7	14.8	28.9	37.7	17.7

TABLE VI.

EXPERIMENTS ON *Chilomonas*.

Table showing results obtained in feeding *Dileptus* on *Chilomonas*. In the five columns under the headings 0-4, are indicated the number of individuals which formed 0, 1, 2, 3, 4, or more vacuoles containing *Chilomonas* in twenty minutes. *Chilomonas* was ingested by 98 per cent. of the dilepti tested and 93 per cent. formed more than one vacuole.

Experiment Number.	Total Number of Dilepti.	Number of Vacuoles Containing <i>Chilomonas</i> .				
		0	1	2	3	4 or More.
1.....	14	0	1	3	4	6
2.....	15	0	0	6	4	5
3.....	15	0	1	3	4	7
4.....	15	0	1	4	2	8
5.....	15	0	0	2	7	6
6.....	15	1	1	5	3	5
7.....	16	1	0	3	4	8
8.....	15	0	2	3	7	3
9.....	15	0	2	1	4	8
10.....	15	0	0	4	3	8
Average.....	15	.2	.8	3.4	4.2	6.4
Per cent.....		1.3	5.3	22.7	28	42.7

By comparing these results with those on inanimate substances as shown in Tables I., II., and III. it will be noted at once that the living material was ingested by a much greater percentage of the dilepti tested, and also that the average number of vacuoles formed during the twenty minutes of the experiment was almost three times as great in the experiments on living material as in those on inanimate substances. It will also be noted that when feeding on inanimate substances *Dileptus* tends to stop feeding after having formed one vacuole, but that when it is feeding on animate substances it does not. This clearly indicates some power of selection, for *Dileptus* refuses to take in useless materials while it ingests nutritive substances in large amounts.

Having thus observed that *Dileptus* can select between animate and inanimate substances, the question naturally arose as to whether there is any choice between different kinds of organisms. Many species of organisms were used in attempting to answer this question. The same methods were used as described above, but only the results obtained in observations on the actual process of feeding were recorded.

The results obtained in these observation, in so far as they pertain to the problem of selection, are briefly summarized in Table VII. By referring to this table it will be seen that *Dileptus* does

TABLE VII.

DISCRIMINATION BETWEEN DIFFERENT ORGANISMS.

Table giving results of feeding tests with *Dileptus* showing selection among living organisms.

I. Organisms	II. Organisms	III. Organisms Never Captured or Injured in Any Way
Readily Captured.	Captured only Rarely.	<i>Paramecium caudatum</i>
<i>Euglena gracilis</i> (?)	<i>Paramecium aurelia</i>	<i>Frontonia</i> ¹
<i>Trachelmonas</i>	Rotifers	<i>Euplotes</i>
<i>Amaba</i>	<i>Stylonychia</i>	<i>Nassula</i>
<i>Halteria</i>	<i>Spirostomum</i>	
<i>Urocetrum turbo</i>		
<i>Chlomonas paramecium</i>		
<i>Colpidium</i>		
<i>Colpoda</i>		
<i>Stentor caruleus</i>		
<i>Stentor polymorphus</i>		

¹Two races of *Frontonia* were used, the one being entirely immune to the attacks of *Dileptus*, and the other only partially.

not capture all organisms, but selects from among the different kinds of living organisms in accordance with the grouping shown. The basis for this grouping will be more readily understood after presenting a few observations on the mechanics of feeding and the function of the trichocysts.

4. OBSERVATIONS ON THE MECHANISM OF FEEDING.

Many detailed observations were made on the mechanics of the feeding process in *Dileptus* in the hope of ascertaining the nature of the power of choice which *Dileptus* has been shown to possess. A few of them, illustrating the various factors involved in the feeding process, are described below.

A. *Euglena*.

In making observations on the capture and ingestion of *Euglena*, a single starved dileptus was isolated in a minute drop of water on a glass slide. To this another small drop containing many euglenæ was added. The reactions were observed under a magnification of about 350 diameters. In numerous observations it was found that whenever a euglena came in contact with any part of the oral surface of the proboscis of the dileptus, it at once become motionless (Fig. 2, a), and remained so for a longer or shorter

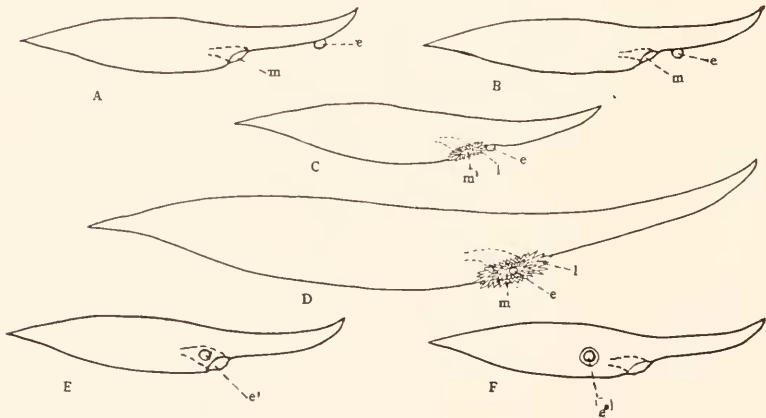


FIG. 2. Diagrammatic sketches illustrating the process of feeding. A-F, successive stages in process of ingesting *Euglena*. *e*, euglena paralyzed by trichocysts; *m*, mouth; *m'*, mouth with protruding lips (*l*); *e'*, euglena being engulfed; *e''*, euglena in food vacuole.

period, depending upon various conditions. It then reacted negatively with great vigor, and simultaneously or occasionally after a very brief latent period, it bulged out in the center and went through the typical euglenoid contractions with great vigor, sometimes holding the contracted form for two or three minutes. If, immediately after the first reaction, it failed to get out of the oral current which is continuously produced by the band of cilia on either edge of the flattened surface of the proboscis, it was carried to the mouth and engulfed (Fig. 2, *b*). If, on the contrary, the first reaction carried it outside the influence of the oral current, it began after a short interval to show some activity and soon recovered, unless it again came in contact with the proboscis. If this occurred, it was almost always carried to the oral region and engulfed.

The process of engulfing was quite extraordinary. Whenever the oral current carried a euglena to the mouth, the gullet, apparently owing to mechanical stimulation, protruded so that a mass of viscous protoplasm was exposed (Fig. 2, *c*). If any particle came in contact with this it adhered, and when this occurred the gullet was again drawn in carrying the particle with it (Fig. 2, *d, e*). In this process there was apparently some suction, for considerable water was always taken in with the solid particles (Fig. 2, *f*).

These observations were repeated on many favorable occasions, and, furthermore, are substantiated in the main by a similar observation recorded by Wrzesniowski ('70). Referring to the capture of a *Stylonychia* by *Dileptus*, Wrzesniowski says, "it tries by means of its proboscis to bring it down into its occasionally wide open mouth, whereupon the *protruding lips* seize so firmly upon the captured little animal that the latter is bitten in two." My observations agree with only the first part of this quotation; the idea of biting is quite contrary to the results of any observations which I have made.

B. ROTIFERS.

In the observations on feeding on rotifers described below, nine specimens were added to a small amount of water containing about twenty starved dilepti. Within two minutes all the rotifers were attacked. The dilepti appeared to sense (?) the rotifers while still

at a distance at least equal to their own length. Sometimes as many as two or three of these ciliates were seen to gather around and attack a single rotifer. In these attacks the dilepti usually failed to capture the rotifers. Only in one instance was a rotifer actually observed to be captured and eaten. Although the feeding process rarely culminated successfully, this experiment afforded observations which are very instructive, as the following indicate.

In these observations each dileptus was continuously swinging its proboscis back and forth, and at the same time revolving on its longitudinal axis. Thus it struck the rotifers, now with the aboral side, now with the oral side of the proboscis, and the corresponding differences in the reactions of the rotifers were most striking. The rotifer in question is one which attaches itself quite securely to the wall of the dish. It also elongates and contracts from time to time without changing its location. When the aboral side of

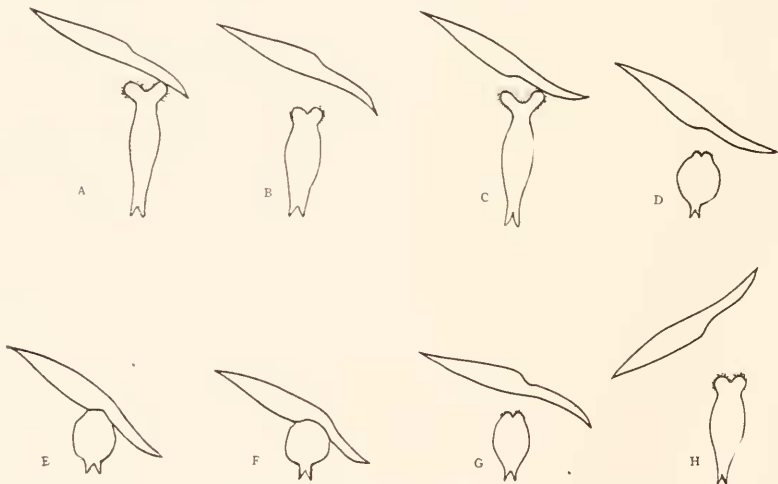


FIG. 3. Sketches illustrating the effect of *Dileptus* on rotifers. A-H, successive stages. Note differential effect, as seen by degree of subsequent contraction, of contact with aboral surface of proboscis as seen in A and B, and of contact with oral surface of proboscis as seen in C and D. For further explanation see text.

the proboscis of a dileptus came in contact with an elongated rotifer it contracted but slightly (Fig. 3, B), if at all, but when the oral surface of the proboscis struck the rotifer it contracted completely

and vigorously, and often remained thus contracted for some little time (Fig. 3, *D*). The hungry little ciliates still persisted and an occasional individual was sometimes seen to succeed in getting the head of an attached rotifer far into its oral opening (Fig. 3, *F*), but a sudden contraction on the part of the attached rotifer invariably resulted in freeing the captive.

Thus when a rotifer comes in contact with the oral surface of the proboscis of a dileptus its reaction is extremely vigorous, while if the aboral surface of the proboscis touches the same rotifer little, if any, reaction is observed. The reaction resulting from contact with the aboral surface of the proboscis is just such as would be expected from a slight mechanical stimulus, but the violent reaction observed whenever the oral surface comes in contact with the rotifer is clearly of an entirely different nature. This difference must be in some way related to the difference between the oral and aboral surfaces of the proboscis. The essential difference between these two is the fact that the former contains trichocysts, while the latter does not. The difference in the reaction is probably, therefore, related to the action of the trichocysts.

C. COLPIDIUM.

The observations on *Colpidium*, like those on *Euglena* described above, were made under high magnification. In one of the many experiments two starved dilepti were isolated in a single drop of water and a smaller drop containing numerous specimens of *Colpidium* was added. The latter were so numerous that they were continually coming in contact with various parts of the dilepti. Some, consequently, frequently came in contact with the oral surface of the proboscis, as well as with various regions of the surface of the body. It was very apparent that those which came in contact with the oral surface of the proboscis were the only ones seriously affected. Whenever a *Colpidium* came in contact with this surface of the proboscis it at once became motionless and remained so for a very brief interval (Fig. 4, *A, a*). Then it suddenly became very active and swam away rapidly. Very often, however, with only a part of its body, for the part which came in contact with the proboscis bulged out and seemed to increase in volume, somewhat comparable to that which takes place when water

is added to gelatine, but very much more rapidly. This mass was usually constricted off from the remaining part (Fig. 4, *B*, *b*), sometimes immediately, sometimes later. If this occurred immediately, the portion constricted off was carried to the gullet by ciliary currents and engulfed. Otherwise this portion was dragged along for a longer or shorter period by the active portion of the *Colpidium* (Fig. 4, *c*, *e*). Most of the *Colpidia* which were injured in this way soon died. Only a few of those which were isolated survived, and these had apparently lost only a small portion of their cytoplasm.

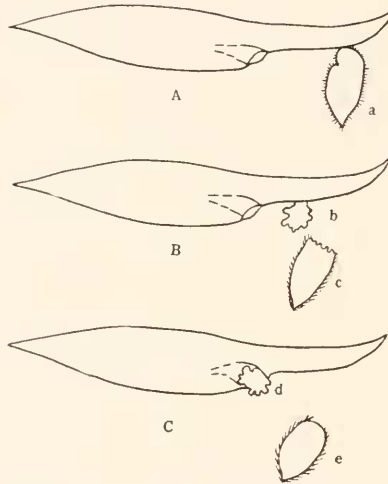


FIG. 4. Sketches illustrating the effects of the trichocysts of *Dileptus* on the infusorian *Colpidium*. *A*, *B*, and *C*, successive stages in the process of feeding. *a*, colpidium (motionless after contact with oral surface of proboscis); *b*, portion of colpidium (apparently cytolized and separated from major portion, *c*); *d*, cytolized portion being engulfed by dileptus; *e*, major portion of colpidium swimming rapidly away.

It seems evident from many observations like the above that it is the trichocysts of *Dileptus* which function primarily in the capture of *Colpidium*. The colpidia are first momentarily paralyzed and then excited to vigorous reaction, but as a rule only after a part of their cytoplasm has been in some way altered so that it disintegrates and *Dileptus* is enabled to feed on it in the same way that it does on other non-motile material.

The results presented seem to indicate that in *Dileptus* there are

two distinct processes involved in feeding: (1) The capture of food and (2) the ingestion of food. In feeding on motile organisms both processes are involved, but when feeding on non-motile substances only the latter process is involved. When feeding on motile forms the prey is first paralyzed, and in many cases where this is only temporary the cytoplasm of the prey is locally seriously affected. In some forms that part of the protoplasm affected is constricted off, while in other forms the entire organism is affected. In this non-motile condition the prey is carried passively by the ciliary current to the buccal cavity and there ingested as described above. The trichocysts are evidently the structures which enable *Dileptus* to capture living prey and make the feeding process of this organism so complicated. The remaining part of the paper will be devoted to observations and discussion as to their nature and function.

5. TRICHOCYSTS.

The nature and function of trichocysts has long been a debated question and even today most authors admit that we know very little about them except in the case of one or two organisms which have been studied very extensively, and even here there is much controversy.

Mitrophanow ('04) maintains that the trichocyst consists of a viscid fluid contained in a cavity in the ectoplasm, whence it is expelled by a sudden contraction of the ectoplasm and stiffens to form a solid thread under the action of the water medium.

Schuberg ('05), however, denies this and maintains that the unexploded trichocyst is a spindle-shaped body with a fine hair-like process at its outer end which reaches to the pellicle, and that when it explodes this material forms into a fine thread-like sharp-pointed rod, often showing a cap-like swelling at one end.

Calkins in 1901 maintained that there are only two types which have been definitely made out—a rod-like form as in *Loxophyllum* and a spindle-shaped form as in *Paramecium*. He also stated that "when protruded from the body they are apparently of the same size and shape as when within the ectoplasm." In 1910, however, the same author wrote concerning trichocysts in general (p. 27), "when the organism is irritated the contents of the capsules are

thrown out with considerable force and the poison which they contain is strong enough to paralyze any single-celled opponent."

Minchin ('12) maintained that "the nature and mechanism of (the peculiar) trichocysts still remains to be explained," but described as typical those forms found in *Paramecium* and *Frontonia*.

Concerning the function of trichocysts there seems to be even less known than there is about the structure. Jennings ('06) wrote that trichocysts "are usually supposed to be weapons of defense, but whether they really serve for defense seems questionable," and suggested that their discharge may be only an expression of injury—"a purely secondary, even pathological phenomenon, like the formation of vesicles on the surface of an injured specimen."

Mast ('09), however, showed clearly that in *Paramecium* the trichocysts have a definite protective function. He observed that the trichocysts of *Paramecium* are discharged in response to injury, produced by *Didinium*, and that as soon as these trichocysts come in contact with the water they form a mass having a firm jelly-like consistency which serves to force the enemy back mechanically, and frequently results in setting the victim free. Calkins ('10, p. 27) says that "sometimes they are used as weapons of offense as well as protective organs," and in another place he describes predaceous protozoa as "usually armed with offensive organs in the form of trichocysts which may be shot out from the surface of the body or carried javelin-like at the extremities of projectile tentacles."

A. OBSERVATIONS ON THE NATURE AND THE FUNCTION OF THE TRICHO CYSTS OF *Dileptus*.

Numerous experiments and observations were made on *Dileptus* to ascertain, if possible, the function as well as the structure of the trichocysts, all of which, as previously stated, are located on the oral surface of the proboscis. A description of a few of the more illuminating of these experiments will follow, but before considering these we may briefly recall a few of the results of the observations on feeding which have a bearing on this subject.

Euglena, it will be recalled, is paralyzed as soon as it comes in contact with the oral surface of the proboscis, and after a short

latent period shows characteristic signs of injury. The violent contraction of the rotifers on every occasion when they come in contact with that portion of *Dileptus* provided with trichocysts gives definite signs of their effect. The observations on *Colpidium* show that the trichocysts not only paralyze this organism, but produce a cytolytic effect upon the protoplasm of the prey.

The following observations are presented in order to show more specifically the precise manner in which these trichocysts function.

a. Effect of Trichocysts on Paramecium bursaria.

In making observations on the action of the trichocysts of *Dileptus* on *Paramecium bursaria*, a single starved dileptus was isolated and added to a small drop of water on a slide containing four specimens of *Paramecium bursaria*. Nearly all the water was then drawn off, after which a cover-glass ringed with vaseline was applied. Two of the paramecia were lost, but the remaining two and the dileptus were confined in so small an amount of water and were so much compressed that they could move only very slowly, and never more than their own length from the others. Consequently all reactions could be observed very accurately. The dileptus, although so compressed that it was more than three times its normal width, continued to rotate on its longitudinal axis and its proboscis was consequently thrown from one side to the other. On several occasions the posterior end of the dileptus came in contact with one of the paramecia, making small indentations in it without any noticeable reaction on the part of the latter. When, however, it slowly reversed its position and the oral surface of the proboscis came in contact with the paramecium, a sudden discharge of trichocysts from the paramecium was observed, so dense as to force the dileptus away. The latter continued to rotate slowly, all the time removing the barrier of trichocysts by means of its ciliary action. The next time only the aboral surface of the proboscis came in contact with the paramecium and no reaction resulted. The third time the proboscis struck the paramecium it was at a slightly different spot and another discharge of trichocysts resulted from the latter. After some little time this again was cleared away, and a fourth attack occurred at about the same spot as the first, this time with an entirely different result. The paramecium

reacted much more violently than previously and at the point of contact a noticeable bulging of the protoplasm occurred. The next attack was at a new spot, with the characteristic discharge of trichocysts. But the following one was at approximately the same spot as the preceding. The protoplasm this time bulged out and formed a large protuberance, even some of the zoöchlorellæ flowing out into it. After half an hour four such protuberances were observed, in all of which it was evident that the pellicle of the paramecium had given way at one small spot, and that the mass of protoplasm which flowed out formed a protuberance with only a narrow connection with the interior. After repeated attacks the paramecium disintegrated.

The other paramecium, meanwhile, had slowly moved toward the dileptus, and this afforded an opportunity to repeat the observations just described. The results obtained in these observations were essentially the same as those obtained in the first observation. The second paramecium, however, appeared to react more vigorously and it consequently escaped more of the attacks than the first, with the result that at the end of half an hour it had only two protuberances, whereas the first, as previously stated, had four. Apparently each attack on the part of the dileptus was just as powerful at the end of the experiment as it was at the beginning.

b. Effect of Trichocysts on Stentor cæruleus.

Perhaps the most instructive, at least the most spectacular, experiment concerning the action of the trichocysts of *Dileptus* is one which can be performed very simply as follows: A dozen or more dilepti are starved for two days; a large blue stentor is then introduced, and the scene of a veritable barbecue is soon presented. The dilepti collect about the stentor and can be seen to strike the latter with their proboscides (Fig. 5). The surface that comes in contact with the stentors in this reaction does not appear to be purely accidental, for it was observed that the oral surface of the proboscis came in contact almost without exception. At the point of contact the pellicle of the stentor gives way momentarily (Fig. 5, *D, d*), and a globular mass of protoplasm is extruded. This mass is soon constricted off (Fig. 5, *F, b*), and the wound apparently heals over at once, while the extruded protoplasmic mass is

readily ingested by the dileptus (Fig. 5, *G, b*). Thus, now here, now there, the stentor gives up part of its protoplasm and each part is eaten by the little dilepti, which sometimes more than treble in size after feeding on this organism. Meanwhile the stentor

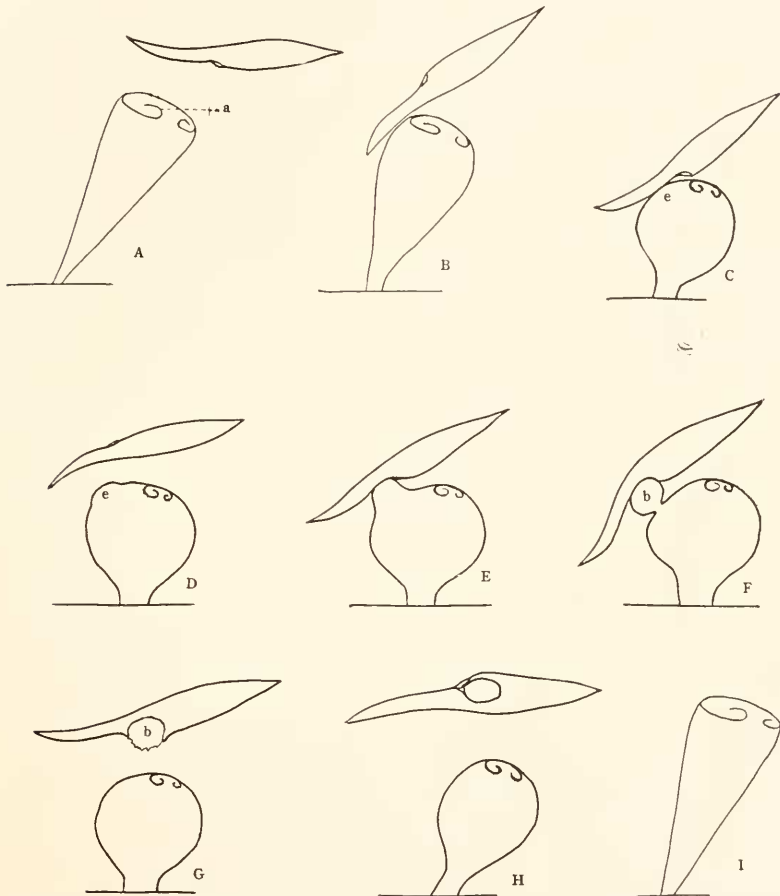


FIG. 5. Sketches illustrating effects of trichocysts of *Dileptus* on *Stentor caruleus*. *A-I*, successive stages. *A*, normal stentor with expanded peristome (*a*). When stimulated as by contact with aboral surface of dileptus, the stentor contracts slightly as shown in *B*. If the oral surface of the proboscis comes in contact (*C*) the stentor contracts vigorously and its protoplasm in the region of contact (*e*) soon protrudes, *D, E*, and *F*. The injured area (*e*) soon gels and the protruding mass (*b*) is constricted off, frequently being ingested by the dileptus (*G*). The stentor remains contracted for some time, but eventually, unless repeatedly attacked, it expands completely (*I*) and appears to be entirely normal.

continually shrinks in size, and unless it escapes before it is too badly injured it dies.

The trichocysts of *Dileptus* evidently affect the ectoplasm of *Stentor* and, as in the case of *Colpidium* previously described, result in a cytolytic action on the surface of the prey at the point of contact. This results in an outflowing of the inner protoplasm until the injured surface can again gelate in some manner, resulting in a new "pellicle."

If the observations just described are made under high magnification, it can be seen that the proboscis of *dileptus* never comes in actual contact with the body of the *stentor*. They are always separated by a space, at least equal to the sum of the lengths of their respective cilia. This would lead to the conclusion that the trichocysts of *Dileptus* are discharged through some little distance—that is, they are thrown out with some force.

c. Effect of Trichocysts on Paramecium aurelia.

Dileptus is normally unable to injure *Paramecium aurelia* in any way, but in one experiment several paramecia were seriously injured by two *dilepti*. In two instances, which were carefully observed, the paramecia appeared to be completely paralyzed, although only momentarily, immediately upon coming in contact with the oral surface of the proboscis. When the proboscis touched the paramecia they reacted vigorously and swam away, but not before they were injured. It was observed that they became much deformed soon after the attack, doubling on the point that had been injured to such an extent that they assumed the form of a horse shoe. One of these paramecia was attacked a second time while in this semi-quiescent condition and was successfully engulfed. The other one was isolated on a hollow ground slide and after about an hour began to swim about, gradually losing its deformity. On the following day it appeared to be normal. The ectoplasmic pellicle of *Paramecium aurelia* is probably of such a nature that it prevents any cytolytic action resulting from the trichocysts. The injurious effect of these structures seems to be due to the production of a definite wound at the point of contact.

d. Effect of Trichocysts on Spirostomum.

When a spirostomum is attacked by a dileptus it contracts vigorously as soon as "stung" (Fig. 6, *A, B*). This usually produces a violent reaction which serves to get it out of reach of the dileptus

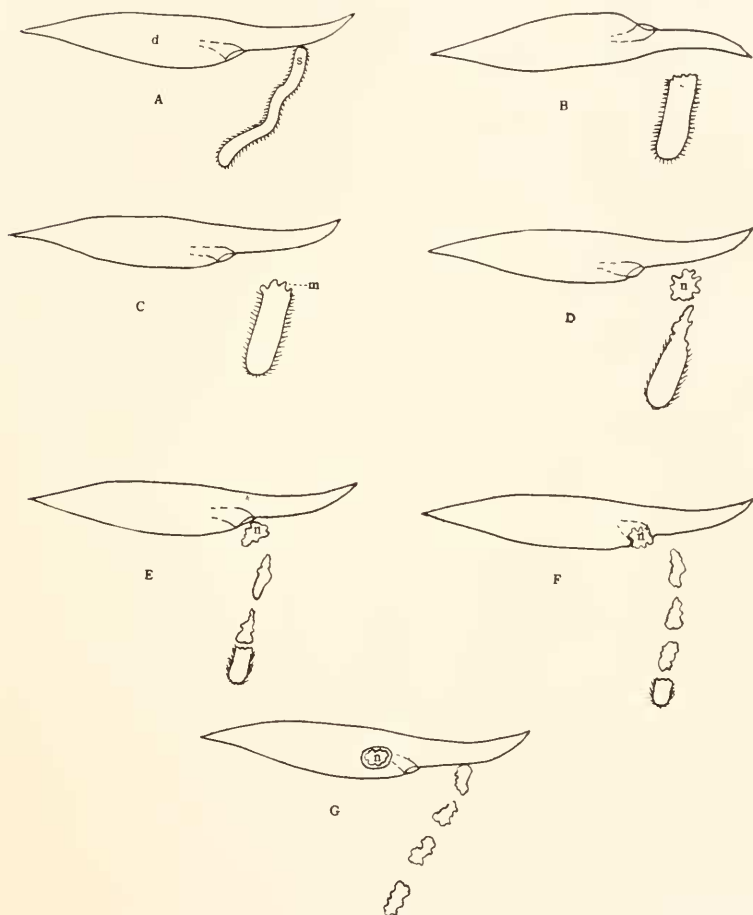


FIG. 6. Sketches illustrating the effect of trichocysts of *Dileptus* on *Spirostomum*. *A-G*, successive stages in process of feeding. When dileptus (*d*) comes in contact with spirostomum (*s*) as in *A*, the latter contracts vigorously and remains momentarily motionless (*B*). Cytolysis begins at area of contact (*m*) and as the spirostomum reacts negatively, swimming rapidly away, the cytolytic process continues, as in *D, E*, and *F*. Meanwhile dileptus has engulfed one or more masses of the disintegrating spirostomum (*n*) as shown in *D-G*.

and thus prevents a second attack. If it is attacked a second time, the result is extensive disintegration of the protoplasm around the point of contact (Fig. 6, *C, m*). If this point is located at one end of the spirostomum, the opposite end swims rapidly away, leaving behind a trail of disintegrating protoplasm (Fig. 6, *E-G*). When any part of the protoplasm of this organism begins to disintegrate, the cytolysis, once begun, progresses rather rapidly until the whole organism has disintegrated. This effect is in contrast with that obtained in *Stentor*, in which an attack produced only local and partial disintegration. Apparently the protoplasm of *Spirostomum* does not possess the power of gelation as observed in *Stentor*, and thus the cytolytic action continues until the organism is disintegrated (Fig. 6, *G*).

The results obtained in numerous other observations made on various other organisms are all in harmony with those which have been described. All these observations seem to show conclusively that the trichocysts discharged by *Dileptus gigas*, first temporarily paralyze the prey, then produce a period of increased activity in the nature of a negative reaction on the part of the prey, and simultaneously effect a cytolytic action at the point of contact.

B. OBSERVATIONS ON THE STRUCTURE OF THE TRICHOCYSTS OF *Dileptus*.

Numerous specimens of *Dileptus* were fixed during various stages in the process of feeding and many different methods of fixation and subsequently staining were employed in an attempt to ascertain the structure of the trichocysts. Before they are discharged the trichocysts can be clearly seen in all properly stained specimens (Fig. 7). They are found, as previously stated, in a band on the oral surface of the proboscis. When stained they appear as elongated bodies (Fig. 7, *t*), which do not show any definite internal morphological structure, but appear to be more or less granular, and if stained at all, are always stained deeply. Fig. 7 shows the relative number, size, and shape of the trichocysts as seen in 4μ sections (*A-D*), and in a total mount (*E*). In favorable specimens they can be seen in the living animal, where they appear as colorless, rather transparent bodies which change shape as the animal twists and turns. They have been seen to become

almost spherical in shape when under pressure from the cover-glass.

In all my work it has been impossible to observe any structures or formed elements of any kind which could be identified as trichocysts or their contents outside the body of *Dileptus*. The safranin

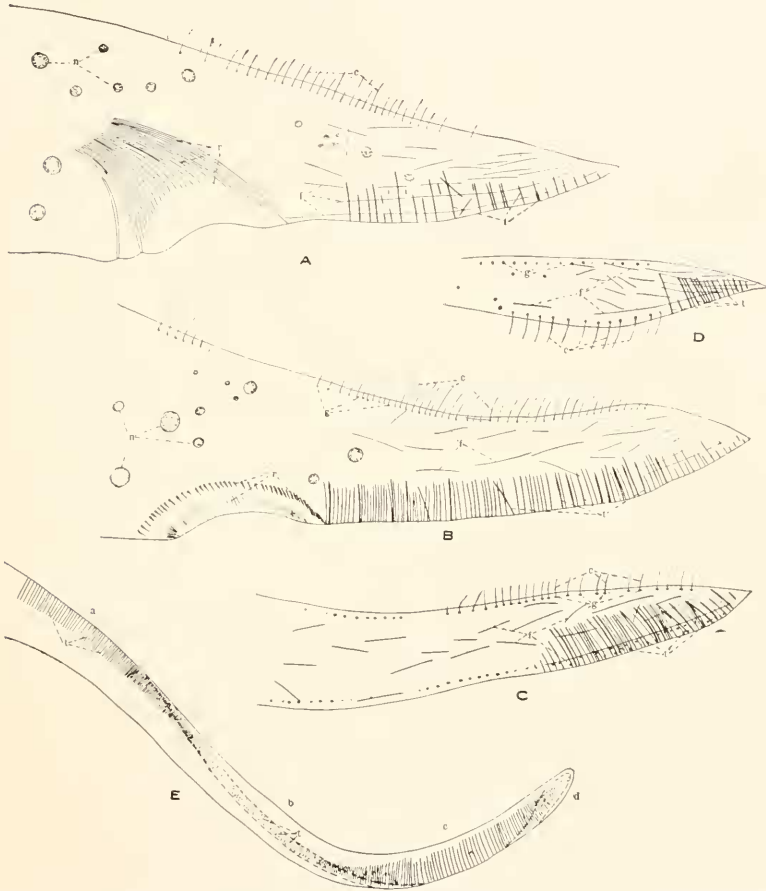


FIG. 7. Camera lucida drawings from preparations fixed in Schaudinn's fluid and stained with Fe-Haem. (A-D) or with acid borax carmine (E). A, B, C, and D are 4μ serial sections of a proboscis. Note size, shape, and number of trichocysts (*t*); the pharyngeal rods (*r*); cilia (*c*); basal granules (*g*) "distributed nucleus," some of which are designated as *n*; and the contractile fibrillae (*f*). E, proboscis of dileptus, slightly twisted; from an entire mount. Note relative number and position of trichocysts (*t*), forming a band on the oral surface of the proboscis. Due to the turn in the proboscis, the trichocysts are seen from side view at *a* and *c* and from end view at *b* and *d*.

method of staining "intra vitam," which demonstrates so clearly the poisonous threads of the nematocysts of the common fresh-water hydra, was also tried, but gave only negative results.

We would maintain, then, that the contents of the trichocysts of this organism do not have a morphological structure after they are discharged, such as the trichocysts of *Paramecium* and *Frontonia* have. It would appear that the trichocysts of *Dileptus* are more like elongated sacs of toxic fluid, which collapse upon discharging the fluid.

If it is true that these structures found in the proboscis of *Dileptus* are bags filled with a poisonous fluid, it is evident that the term trichocyst (hair-sack) is not exactly applicable, and in order to be more exact the term toxicyst (poison-sack) might be employed.

C. SUMMARY OF OBSERVATIONS ON TRICHO CYSTS.

The foregoing observations and experiments show that the trichocysts of *Dileptus* are the structures which this organism employs in capturing food. They have the power to paralyze some organisms, to bring about the cytolysis of others, and to cause a vigorous reaction in almost all infusoria. Organisms like *Paramecium* and *Frontonia* are probably protected against the ordinary attacks of *Dileptus* by their own protective trichocysts. Organisms like *Euplotes*, which are provided with a lorica, form another class of infusorians which appear to be protected against the trichocysts of *Dileptus*. Certain species of *Stylo nychia* are known to possess a heavy cuticle resembling a lorica, and it is perhaps for this reason that these organisms were but rarely observed to fall prey to *Dileptus*. The great majority of ciliates seem to fall prey to *Dileptus*, either owing to the paralyzing effect of its trichocysts or to the cytolytic action of these structures.

6. THE MECHANISM OF SELECTION OF FOOD IN *DILEPTUS*.

There appear to be two distinct mechanisms by which selection of food is brought about in *Dileptus*. (1) The rejection of inorganic particles, as shown in Tables I., II., and III., is evidently due to the effect of the physiological state, which serves to prevent the organism from ingesting more. (2) The purely chemical and

physical properties of the trichocysts of *Dileptus* seem to determine very largely the nature of the food which this organism ingests. If the trichocysts are able to bring about cytolysis of the protoplasm of an organism, or even to completely paralyze it for a time, that organism is "selected" as food. This relation between the protoplasm of the prey and the trichocysts of *Dileptus* is the important factor in determining whether or not *Dileptus* "selects" it as food.

In *Dileptus* the former mechanism seems to play but a small rôle. Because of its natural habits this ciliate deals almost exclusively with living organisms. As previously stated, *Dileptus* thrives only in "relatively pure" and quiet water in which there are but few inorganic particles in suspension. Its habit of continually swimming serves admirably to keep it off the substratum, and we can readily comprehend that motile organisms are almost the only substances from which it has normally to select. We can safely conclude that much of the power of selection of food in *Dileptus* resides in the peculiar properties of its trichocysts.

7. SUMMARY.

1. *Dileptus gigas* normally feeds on living organisms, but under certain conditions it ingests inanimate particles.

2. It discriminates between living organisms and inanimate substances, ingesting the former in large amounts, while the latter are only sparingly ingested.

3. *Dileptus* selects from among different kinds of organisms, eating some with great readiness, while others are rarely ingested.

4. It captures its prey by means of trichocysts which either paralyze the prey, *e.g.*, *Euglena*, or bring about cytolysis of all or part of the protoplasm of the prey, *e.g.*, *Colpidium* and *Stentor*.

5. The trichocysts are probably of a liquid nature, highly toxic, with specific cytolytic properties.

6. The trichocysts of *Dileptus* are used for the purpose of capturing food.

7. Selection of food in *Dileptus* depends on two factors: (*a*) The physiological state of the organism itself, which appears to determine whether a substance shall be ingested in large or small amounts, and (*b*) the chemical properties of its trichocysts, which

determine in large measure whether any living organism can or can not be successfully captured.

8. Specialized structures as, for example, the trichocysts of *Paramecium* and the lorica of *Euplotes*, serve as protection against the attacks of *Dileptus*.

8. LITERATURE CITED.

Bütschli, O.

'80-89 Bronn's Tierreichs, Bd. I., Protozoa. Leipzig. 2035 s.

Calkins, G. N.

'01 The Protozoa. New York. 347 pp.

'09 Protozoölogy. New York. 349 pp.

Claparède and Lachman.

'85 Sur la reproduction des Infusoires. Ann. des. Sci. Nat., ser. 4, tome 8, pp. 221.

Ehrenberg, C. G.

'38 Die Infusionsthierchen als Vollkommene Organismem. Leipzig.

Entz, G.

'88 Studien über Protisten. Aufträge der Königl. Ung. Naturw. Ges., Budapest.

Hausman, L. A.

'17 Observations on the Ecology of the Protozoa. Am. Nat., Vol. 51, pp. 157-172.

Hertwig, R.

'02 Nuclear Cytoplasmic Relations. Sitz d. Ges. f. Morph. u. Phys., Bd. 18, s. 77-100.

'04 Conjugation in *Dileptus gigas*. Sitz. d. Ges. f. Morph. u. Phys., Bd. 20, s. 1-4.

Jennings, H. S., and Moore, E. M.

'02 Studies on Reactions to Stimuli in Unicellular Organisms. Am. Jour. of Psychology, Vol. 6, pp. 223-250.

Jennings, H. S.

'06 The Behavior of the Lower Organisms. New York. 336 pp.

Khainsky, A.

'10 Morphologie und Physiologie einiger Infusorien. Arch. f. Protistenk., Bd. 21, s. 1-60.

Kent, W. S.

'81 A Manual of the Infusoria. London. 3 vols., 894 pp., 52 pl.

Mast, S. O.

'09 Reactions of *Didinium nasutum* with special reference to the Feeding Habits and the Function of Trichocysts. BIOL. BULL., Vol. 14, pp. 91-118.

Metalnikow, S.

'12 Contributions a l'etude de la digestion intra-cellulaire. Arch. de. Zool. Exper., tome 49, ser. 4.

Minchin, E. A.

'12 Introduction to a Study of the Protozoa. London. 520 pp.

Mitrophanow, P.

- '04 La Structure (etc.) des Trichocysts des Paramecies. Arch. f. Protistenk., Bd. 5, s. 78-92.

Moody, J. E.

- '12 Observations on the Life History of Two Rare Ciliates, *Spathidium spathula* and *Actinobolus radians*. Jr. Morph., Vol. 23, pp. 349-407.

Pritchard, A.

- '61 Infusoria. London. 968 pp.

Root, F. M.

- '14 Reproduction and Reactions to Food in the Suctorian, *Podophrya colini* n. sp. Archiv. f. Protistenk., Bd. 35, s. 164-197.

Schaeffer, A. A.

- '10 Selection of Food in *Stentor*. Jour. Exper. Zool., Vol. 8, pp. 75-132.
'16 On the Feeding Habits of *Amaba*. Jour. Exper. Zool., Vol. 20, pp. 529-584.

Schuberg, A.

- '05 Über Cilien und Trichocysten einiger Infusorien. Arch. f. Protistenk., Bd. 6, s. 61-111.

Verworn, M.

- '89 Psychophysiologische Protistenstudien. Jena. s. 218.

Wrzesniowski, A.

- '70 Infusorien von Warschau. Zeit. Wiss. Zool., Bd. 20, s. 467-512.